

(19) World Intellectual Property
Organization
International Bureau



(43) International Publication Date
23 October 2003 (23.10.2003)

PCT

(10) International Publication Number
WO 2003/087367 A3

(51) International Patent Classification⁷: C12N 15/11,
A61K 31/713, C12Q 1/68, C12N 15/86, 5/10, A01K
67/027

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(21) International Application Number:
PCT/EP2003/004002

(22) International Filing Date: 16 April 2003 (16.04.2003)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
02008761.5 18 April 2002 (18.04.2002) EP
60/431,173 5 December 2002 (05.12.2002) US

(81) Designated States (*national*): AE, AG, AL, AM, AT, AU,
AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU,
CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC,
LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW,
MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE,
SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ,
VC, VN, YU, ZA, ZM, ZW.

(84) Designated States (*regional*): ARIPO patent (GH, GM,
KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW),
Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM),
European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE,
ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO,
SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM,
GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

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Published:
— *with international search report*

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(88) Date of publication of the international search report:
29 April 2004

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: MEANS AND METHODS FOR THE SPECIFIC INHIBITION OF GENES IN CELLS AND TISSUE OF THE CNS AND/OR EYE

(57) Abstract: Described is a method for the specific modulation of the expression of target genes in cells and/or tissues of the CNS and/or eye, wherein a composition comprising one or more doubled stranded oligoribonucleotides (dsRNA) is introduced into the cell, tissue or organism outside the blood-brain or blood-retina barriers. Furthermore, a method for the identification and validation of the function of a gene is provided, wherein the method provides a test cell, test tissue or test organism, which allow information to be gained on the function of the target gene. In addition, compositions and kits are described useful for those methods. In particular, components and methods for the diagnostic use and/or therapy of disorders related to the CNS and/or eye are provided which are based on RNA interference.



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INTERNATIONAL SEARCH REPORT

International Application No.
PC 03/04002

A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12N15/11 A61K31/713 C12Q1/68 C12N15/86 C12N5/10
A01K67/027

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12N A61K C12Q A01K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	DZITOEVA S. ET AL.: "Intra-abdominal injection of double-stranded RNA into anesthetized adult <i>Drosophila</i> triggers RNA interference in the central nervous system" MOLECULAR PSYCHIATRY, vol. 6, no. 6, November 2001 (2001-11), pages 665-670, XP008023738 the whole document	1-6, 11-18, 32-34, 41, 42, 44-47
X	GAN L. ET AL.: "Specific interference of gene function by double-stranded RNA in neuronal cell lines" SOCIETY FOR NEUROSCIENCE ABSTRACTS, vol. 27, no. 2, 2001, page 2051 XP002259262 the whole document	2-6, 11-18, 29, 30, 32, 41, 44-47
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☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents:

A document defining the general state of the art which is not considered to be of particular relevance

E earlier document but published on or after the international filing date

L document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

O document referring to an oral disclosure, use, exhibition or other means

P document published prior to the international filing date but later than the priority date claimed

T later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

X document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

Y document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

Z document member of the same patent family

Date of the actual completion of the international search

20 January 2004

Date of mailing of the international search report

10 FEB 2004

Name and mailing address of the ISA

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP 03/04002

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
see FURTHER INFORMATION sheet PCT/ISA/210
2. ☐ Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this International application, as follows:

see additional sheet

1. ☒ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☒ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: 1-6, 11-25, 27-34, 39, 41, 44-47 all partially

Use of one or more double-stranded oligoribonucleotides (dsRNA) for the preparation of a composition for the specific modulation of the expression of one or more target genes in cells and/or tissues of the CNS of a subject, wherein said composition is designed to be applied outside the blood-brain barrier.

A method for the specific modulation of the expression of target genes in cells and/or tissues of the CNS, wherein a composition comprising one or more double-stranded oligoribonucleotides (dsRNA) is introduced into a cell, tissue or organism outside the blood-brain barrier.

Uses, methods, cells, tissues, non-human organisms, diagnostic compositions, kits related thereto.

2. Claims: 7-10, 26, 35-38, 40, 42, 43 all totally; 1-6, 11-25, 27-34, 39, 41, 44-47 all partially

Use of one or more double-stranded oligoribonucleotides (dsRNA) for the preparation of a composition for the specific modulation of the expression of one or more target genes in cells and/or tissues of the eye of a subject, wherein said composition is designed to be applied outside the blood-retina barrier.

A method for the specific modulation of the expression of target genes in cells and/or tissues of the eye, wherein a composition comprising one or more double-stranded oligoribonucleotides (dsRNA) is introduced into a cell, tissue or organism outside the blood-retina barrier.

Uses, methods, cells, tissues, non-human organisms, pharmaceutical and diagnostic compositions, kits related thereto.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.1

Although claims 2-31 and 47 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

Although claims 3-31 and 47 are directed to a diagnostic method practised on the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

INTERNATIONAL SEARCH REPORT

International Application No
PC 03/04002

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	<p>WO 01 82900 A (THE REGENTS OF THE UNIVERSITY OF CALIFORNIA (US); PARDRIGE WILLIAM M.) 8 November 2001 (2001-11-08)</p> <p>the whole document</p>	<p>1-3, 5, 6, 11, 12, 16-22, 24, 25, 27-33, 39, 40, 44, 45, 47</p>
Y	<p>WO 00 63364 A (AMERICAN HOME PRODUCTS CORPORATION; PACHUK CATHERINE SATISHCHANDRAN C.) 26 October 2000 (2000-10-26)</p> <p>page 21 page 39 -page 42; example 3</p>	<p>1-3, 5-12, 16-22, 24-33, 39, 40, 44, 45, 47</p>
Y	<p>WO 01 83729 A (NOVARTIS AG (CH); SCRIPPS RES INST; NEMEROW; VON SEGGERN; FRIEDLANDER) 8 November 2001 (2001-11-08)</p> <p>the whole document page 17, line 15-17</p>	<p>1-3, 5-12, 16-22, 24-33, 39, 40, 44, 45, 47</p>
A	<p>EP 0 308 066 A (HEM RESEARCH, INC. (US); CARTER WILLIAM A.) 22 March 1989 (1989-03-22) page 2, line 40 page 3, line 28-30</p>	<p>1</p>
A	<p>WO 01 75164 A (WHITEHEAD INST; MAX PLANCK; MIT; MASSACHUSSETS UNIV; TUSCHL; SHARP ET) 11 October 2001 (2001-10-11) cited in the application the whole document</p>	<p>13-15</p>
A	<p>WO 97 20579 A (SMITHKLINE BEECHAM PLC (GB); PRICE JACK) 12 June 1997 (1997-06-12) the whole document</p>	<p>21, 23</p>
A	<p>WO 98 22132 A (YISSUM RES DEV CO HEBREW UNIV JERUSALEM; SOREQ FRIEDMAN SEIDMAN KAUFER) 28 May 1998 (1998-05-28) page 15, line 23-26</p>	<p>21, 23</p>
	-/--	

INTERNATIONAL SEARCH REPORT

International Application No

PC 03/04002

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	CARLSON S.D. AND HILGERS S.L.: "Perineurium in the Drosophila (Diptera: Drosophilidae) embryo and its role in the blood-brain/nerve barrier" INTERNATIONAL JOURNAL OF INSECT MORPHOLOGY AND EMBRYOLOGY, vol. 27, no. 2, April 1998 (1998-04), pages 61-66, XP002259381 the whole document	
X	WO 01 36646 A (CANCER RES CAMPAIGN TECH LIM (GB) ZERNICKA-GOETZ; WIANNY; EVANS GLOVER) 25 May 2001 (2001-05-25) page 15 -page 18	1,2,5-7, 11-31, 33,39, 40,44, 45,47
X	WO 00 44914 A (MEDICAL COLLEGE OF GEORGIA RESEARCH INSTITUTE (US); LI; FARRELL; KIRBY) 3 August 2000 (2000-08-03) page 14 page 47 -page 54; claims	1,2,5-7, 11-31, 33,39, 40,44-47
X	PINEDA D. ET AL.: "The genetic network of prototypic planarian eye regeneration is Pax6 independent" DEVELOPMENT, vol. 129, March 2002 (2002-03), pages 1423-1434, XP002259263 the whole document	1-7,11, 12, 15-18, 32,33, 44,45
A	LAWSON S.S. AND JOHNSON C.: "Understanding the Glaucoma gene" MOLECULAR BIOLOGY OF THE CELL. 40TH AMERICAN SOCIETY FOR CELL BIOLOGY ANNUAL MEETING; SAN FRANCISCO, CA, USA; DECEMBER 9-13, 2000, vol. 11, no. supplement , December 2000 (2000-12), page 14a XP008023805 Abstract 69 the whole document	

INTERNATIONAL SEARCH REPORT

Information on patent family members

Patent Application No.
PCT/EP 03/04002

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
WO 0182900	A	08-11-2001	US 6372250 B1	16-04-2002
			AU 5364601 A	12-11-2001
			CA 2403501 A1	08-11-2001
			EP 1282403 A1	12-02-2003
			WO 0182900 A1	08-11-2001
			US 2002054902 A1	09-05-2002
WO 0063364	A	26-10-2000	AU 4472100 A	02-11-2000
			BR 0009884 A	08-01-2002
			CA 2370628 A1	26-10-2000
			CN 1375004 T	16-10-2002
			EP 1171586 A2	16-01-2002
			JP 2002542263 T	10-12-2002
			WO 0063364 A2	26-10-2000
WO 0183729	A	08-11-2001	AU 9519301 A	12-11-2001
			CA 2407881 A1	08-11-2001
			WO 0183729 A2	08-11-2001
			EP 1280929 A2	05-02-2003
			JP 2003531609 T	28-10-2003
			US 2002193327 A1	19-12-2002
EP 0308066	A	22-03-1989	AT 132041 T	15-01-1996
			AU 1001595 A	23-02-1995
			AU 1736792 A	30-07-1992
			AU 2053588 A	20-04-1989
			AU 4849897 A	12-03-1998
			CA 1327002 C	15-02-1994
			CN 1032740 A ,B	10-05-1989
			DE 3854829 D1	08-02-1996
			DE 3854829 T2	14-08-1996
			DK 449888 A	13-02-1989
			EP 0308066 A2	22-03-1989
			FI 883763 A	13-02-1989
			HU 47854 A2	28-04-1989
			IE 72103 B1	12-03-1997
			JP 1131119 A	24-05-1989
			KR 131879 B1	17-04-1998
			NO 883594 A	13-02-1989
			NZ 225799 A	26-04-1991
			OA 8901 A	31-10-1989
			PT 88266 A ,B	30-06-1989
			SU 1834663 A3	15-08-1993
			US 5683986 A	04-11-1997
			ZA 8805967 A	26-07-1989
WO 0175164	A	11-10-2001	AU 3574402 A	11-06-2002
			AU 4962201 A	15-10-2001
			CA 2404890 A1	11-10-2001
			CA 2429814 A1	06-06-2002
			CZ 20031839 A3	15-10-2003
			WO 0244321 A2	06-06-2002
			EP 1309726 A2	14-05-2003
			HU 0302557 A2	28-10-2003
			JP 2003529374 T	07-10-2003
			NO 20032464 A	21-07-2003
			WO 0175164 A2	11-10-2001
			US 2003108923 A1	12-06-2003

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No.
PCT/JP 03/04002

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
WO 0175164	A		US 2002086356 A1	04-07-2002
WO 9720579	A	12-06-1997	WO 9720579 A2	12-06-1997
WO 9822132	A	28-05-1998	AU 732043 B2	12-04-2001
			AU 5364298 A	10-06-1998
			CA 2272280 A1	28-05-1998
			EP 0957932 A2	24-11-1999
			JP 2002512592 T	23-04-2002
			WO 9822132 A1	28-05-1998
			US 6025183 A	15-02-2000
			US 6110742 A	29-08-2000
			US 6258780 B1	10-07-2001
			AU 727611 B2	14-12-2000
			AU 5385698 A	03-07-1998
			EP 0951536 A1	27-10-1999
			JP 2001510336 T	31-07-2001
			WO 9826062 A2	18-06-1998
			US 6121046 A	19-09-2000
WO 0136646	A	25-05-2001	AU 1406501 A	30-05-2001
			CA 2391622 A1	25-05-2001
			DE 1230375 T1	09-01-2003
			EP 1230375 A1	14-08-2002
			WO 0136646 A1	25-05-2001
			JP 2003514533 T	22-04-2003
			NO 20022359 A	18-07-2002
			US 2003027783 A1	06-02-2003
			ZA 200203816 A	02-01-2003
WO 0044914	A	03-08-2000	AU 2634800 A	18-08-2000
			CA 2361201 A1	03-08-2000
			EP 1147204 A1	24-10-2001
			WO 0044914 A1	03-08-2000
			US 2002114784 A1	22-08-2002